Original Article

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Comparison of Ascitic Fluid Cholesterol and Fibronectin with Conventional Ascitic Protein Values in Differentiating Malignant Ascites from Non malignant Ascites: A Cross-sectional Study

Internal Medicine Section

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ABSTRACT

Introduction: It is a known clinical problem to differentiate between malignant and non malignant ascites because there is no single routine biochemical laboratory test that can completely distinguish between them. The diagnostic sensitivity of cytological examination is 40%-60%.

Aim: To establish the correlation and evaluate the levels of ascitic fluid cholesterol and fibronectin in the differentiation of malignant and non malignant ascites, compared to conventional total protein concentration in ascitic fluid.

Materials and Methods: A cross-sectional study included 93 patients with clinically detectable ascites, admitted to the Department of Medicine at SCB Medical College, Cuttack, Odisha, India. Patients over 18 years of age presenting with ascites confirmed clinically or by Ultrasonography (USG) were included. Pregnant patients, those with blunt abdominal injury, those previously diagnosed with cancer and having received anticancer treatment, those who failed to give consent, and critically ill patients were excluded. All patients underwent diagnostic paracentesis, and the ascitic fluid was analysed for gross appearance, cytological examination, and biochemical studies. Statistical analysis was performed using Statistical Package for Social Sciences (SPSS) version 27.0.

Results: The study group comprised 47 males and 46 females. The mean age of the study group was 52.25±12.74 years, ranging from 20 to 79 years. The mean ascitic fibronectin concentration in patients with malignant ascites was 64.93±21.41 ng/mL. Using various cutoff values, the diagnostic accuracy of ascitic fluid cholesterol, fibronectin, total protein, Serum Ascites Albumin Gradient (SAAG), and Serum Ascites Cholesterol Gradient (SACG) in differentiating malignancy-related ascitic fluid from non malignant ascites were determined as 98.92%, 97.85%, 56.99%, 52.69%, and 67.74%, respectively.

Conclusion: Cholesterol and fibronectin estimations present valuable diagnostic features for differentiation, surpassing the conventional protein, albumin, and SAAG determinations in terms of diagnostic accuracy and cost-effectiveness of the assay.

Keywords: Albumin, Carcinoma, Cirrhosis, Serum ascites albumin gradient, Serum ascites cholesterol gradient

INTRODUCTION

Ascites, the pathological accumulation of fluid in the peritoneal cavity, is a frequently encountered situation in clinical practice. It is an important clue to an underlying illness, which may be localised to the peritoneal cavity or secondary to an underlying systemic illness [1]. The primary mechanisms of ascites progression can involve increased hydrostatic pressure (e.g., cirrhosis and congestive heart failure), diminished oncotic pressure (nephrotic syndrome), elevated peritoneal fluid production compared to resorption (neoplasms), or a combination of these factors [2].

The majority of patients presenting with ascites have underlying cirrhosis, while the rest can be due to malignancies, heart failure, tuberculosis, pancreatitis, and other rare causes [3]. Rare causes include constrictive pericarditis, inferior vena cava obstruction, non cirrhotic portal hypertension, portal vein thrombosis, sinusoidal obstruction syndrome, hepatic vein thrombosis, nephrotic syndrome, biliary leak, hypothyroidism, familial Mediterranean fever, etc., [3].

The development of ascites in a patient with cirrhosis suggests progression to a decompensated state and indicates a poor prognosis, as the survival rate significantly decreases [4]. The diagnosis of malignant ascites also carries a grave prognosis, with a survival period of only about 20 weeks without intervention [5]. Treatment depends on the underlying cause. However, differentiating between malignant and non malignant ascites is a known clinical challenge, as there is no single regular biochemical laboratory test that can definitively distinguish between them. Even the cytological examination, while highly specific, has a diagnostic sensitivity of only about 40%-60% [6]. As a result, additional parameters of ascitic fluid have been evaluated for their differential diagnostic significance, such as total protein concentration, which is commonly used. There are no specific distinguishing features, and no particular diagnostic test is accurate in differentiating malignant and non malignant ascites. There is a possibility of false-positive results in cytological examination, as reactive mesothelial cells in the ascitic fluid can mimic malignant cells [6]. Various tumour markers (such as Carbohydrate Antigen 19-9 [CA 19-9], Carcinoembryonic Antigen (CEA), Alpha-Foetoprotein (AFP), CA 125, and CA 15-3) are used to diagnose the primary site of malignancy [7]. However, these markers are too sensitive for diagnosis, and the diagnostic performance of these tumour markers in malignant ascites is inconclusive.

Recent investigations have sparked interest in the surface properties of cancer cells, indicating potential novel markers of malignant effusions. Cholesterol and fibronectin levels have been found to be elevated in malignant ascites [8]. Recent studies have demonstrated an efficiency greater than 90% for differentiating malignant ascites from non malignant ascites based on fibronectin and cholesterol concentrations [9]. Moreover, results for ascitic fluid cholesterol and fibronectin can be obtained in less than three hours after

paracentesis, thereby reducing hospital stay for patients. Although the utility of fibronectin has been examined in our setting among patients with sickle cell disease, malnutrition, and pregnancy [8], its role in differentiating ascites has not been investigated. There are limited internationally acknowledged articles regarding the usefulness of ascitic fluid fibronectin, with many of them focused on Caucasians [8,10-12].

Numerous studies have evaluated the role of cholesterol in the differential diagnosis of ascitic fluid [13-20]. However, most of these studies have focused on Caucasian and African populations, with only a few conducted on Asian subjects [13-20]. The present study was aimed to assess the role of fibronectin and cholesterol in differentiating malignant from non malignant ascites, compared to conventional total protein concentration in ascitic fluid and to detect the levels of ascitic fluid cholesterol and fibronectin in patients with ascites, establish the correlation between ascitic fluid cholesterol and fibronectin levels with malignant and non malignant ascites, and evaluate the diagnostic performance of ascitic fluid cholesterol and fibronectin in differentiation malignant and non malignant ascites compared to conventional total protein concentration. The outcome parameters included sensitivity, specificity, Positive Predictive Value (PPV), Negative Predictive Value (NPV), and diagnostic efficiency.

MATERIALS AND METHODS

A cross-sectional study included 93 patients with clinically detectable ascites who were admitted to the Department of Medicine, SCB Medical College, Cuttack, Odisha, India, from between June 2021 and May 2022. The patients were included in the study after considering the inclusion and exclusion criteria, and they underwent a thorough evaluation after obtaining informed consent and ethical clearance from the Institutional Ethical Committee (IEC application no. 867/ Dated. 11.06.2021) prior to the commencement of the study.

Inclusion criteria: Patients aged >18 years presenting with clinically or ultrasonographically confirmed ascites were included in the study.

Exclusion criteria: Pregnant patients, those with blunt abdominal injury, those diagnosed with cancer who had previously received anticancer treatment (chemotherapy and/or radiotherapy), those who failed to give consent, and critically ill patients were excluded from the study.

Study Procedure

The patients were divided into two groups, group A and group B. Group A consisted of patients with non malignant ascites, while group B consisted of patients with malignant ascites. Group A was further divided into subgroup 1 and subgroup 2. subgroup 1 included patients with ascites presenting with clinical features of liver cirrhosis from various causes, confirmed by histological or USG findings. Subgroup 2 included patients with various etiologies of ascites associated with non malignant diseases except liver cirrhosis. Group B was also divided into subgroup 3, which included patients with ascites secondary to peritoneal carcinomatosis, and subgroup 4, which included patients with malignant diseases and liver involvement but without evidence of peritoneal carcinomatosis. Each patient underwent a comprehensive history-taking, systematic physical examination, and standard laboratory assessments including Complete Blood Count (CBC), Random Blood Sugar (RBS), Renal Function Test (RFT), Liver Function Test (LFT), serum protein and albumin, serum electrolytes, haepatitis B surface antigen, and antibody to haepatitis C. USG of the abdomen and pelvis, digital chest X-ray (PA view), and Electrocardiogram (ECG) were performed for every patient. All patients underwent diagnostic paracentesis, and the ascitic fluid was analysed for gross appearance, total protein, albumin, cholesterol, fibronectin, total cell count, differential count, Gram's stain, Ziehl-Neelsen (ZN) stain, culture, Adenosine Deaminase (ADA), Cartridge Based Nucleic Acid Amplification Test (CBNAAT), and cytology. The SAAG was calculated by subtracting the ascitic fluid albumin from the simultaneously obtained serum albumin. The SACG was calculated by subtracting the ascitic fluid cholesterol from the simultaneously obtained serum cholesterol. Ascitic fluid measurements of glucose, amylase, Lactate Dehydrogenase (LDH), and triglycerides were performed only in relevant situations. Fibronectin was quantitatively determined using an Enzyme-linked Immunosorbent Assay (ELISA) kit, Human FN (Fibronectin) ELISA Kit, from ELK Biotechnology. Special investigations such as Computed Tomography (CT) scans of the abdomen and pelvis, colonoscopy, and Two Dimensional (2D) echocardiography were performed in selected cases.

STATISTICAL ANALYSIS

The statistical analysis was performed using SPSS version 27.0. The mean values with Standard Deviation (SD), median values with range of cholesterol, total protein, albumin, and ascitic fluid fibronectin concentrations in plasma and ascitic fluid were calculated. Variance, correlation, and regression analyses were conducted. Receiver Operating Characteristic (ROC) curves were calculated using R-project software by plotting the fraction of true positive rate (sensitivity) against the False Positive Rate (FPR) (1-specificity). The Area Under the Curve (AUC), which is a relative measure of diagnostic test performance, was determined. By overlaying the ROC curves of different markers of malignancy, the most predictive marker could be selected. Cut-off limits for the determined parameters were applied to classify the results into four categories: true positive, true negative, false positive, and false negative.

The sensitivity was calculated as a/a+d×100%, specificity as b/b+c×100%, PPV as a/a+c×100%, NPV as b/b+d×100%, and diagnostic efficiency as (a+b)/a+b+c+d×100%. The significance of differences in sensitivity, specificity, and efficiency among different parameters was assessed using the Chi-square test. A p-value of less than 0.05 was considered statistically significant. The Chi-square test was also used to compare clinical and biochemical characteristics between the levels of fibronectin in malignant and non malignant ascites for discrete variables, and the Student's unpaired t-test was used for continuous variables and applied to compare mean values between groups. The Pearson correlation coefficient was used to assess the correlation between the studied variables. In all tests, a significance level of $p<0.05$ was used.

RESULTS

In the present study, 93 patients with ascites who were attending a Tertiary Care Hospital were included. Out of these, 47 (50.5%) were males and 46 (49.5%) were females. [Table/Fig-1] displays the division of patients into groups and subgroups based on the causes of ascites. A gender difference was also observed between the two groups, with group A having more males and group B having more females. The mean age of the study group was 52.25±12.74 years, ranging from 20 to 79 years. Patients in group B were slightly older than those in group A (mean age±SD: 55.79±12.48 vs 48.63±12.09, respectively). Ascitic fluid cytology was positive in 70.2% of patients with malignancies in the present study. [Table/Fig-2] presents the mean values with SD, as well as median values with range, of ascitic fluid concentrations of cholesterol, fibronectin, protein, and albumin. The dissimilarity in ascitic fluid concentrations was more noticeable for cholesterol and fibronectin compared to protein when patients with malignant ascites and those with non malignant ascites were considered simultaneously.

The mean plasma values of cholesterol, total protein, and albumin were 165.89±45.81 mg/dL, 6.41±1.00 g/dL, and 3.05±0.74 g/dL, respectively. [Table/Fig-3] presents the corresponding values of ascitic fluid fibronectin, cholesterol, total protein, SAAG, and SACG for malignant and non malignant ascites. The mean ascitic fibronectin concentration in patients with malignant ascites was

Type and causes of ascites			
Group A (non malignant ascites) 46 (49.5%) patients (29 males and 17 females)		Group B (malignant ascites) 47 (50.5%) patients (18 males and 29 females)	
Subgroup 1 *(ascites with histologically or ultrasonographic finding proven liver cirrhosis from various causes)* **28 (30.11%) patients (21 males and 7 females)**	**Subgroup 2** *(miscellaneous causes of ascites related to non malignant diseases other than liver cirrhosis)* **18 (19.35%) patients (8 males and 10 females)**	**Subgroup 3** *(ascites secondary to peritoneal carcinomatosis)* **44 (47.31%) patients (17 males and 27 females)**	**Subgroup 4** *(ascites with malignant diseases and affection of the liver, but without evidence of peritoneal carcinomatosis)* **3 (3.23%) patients (1 male and 2 females)**
• Alcoholic in 16 • Chronic viral haepatitis in 5 (haepatitis B in 3 and haepatitis C in 2) • Non alcoholic steatohaepatitis in 2 • Primary biliary cirrhosis in 1 • Autoimmune haepatitis in 1 • Wilson's disease in 1 • Mixed or cryptogenic in 2	• Congestive heart failure in 4 • Chronic kidney disease in 3 • Nephrotic syndrome in 1 • Portal vein thrombosis in 1 • Pancreatitis in 2 • Peritoneal tuberculosis in 5 • Systemic lupus erythematosus in 1 • EHPVO n 1	• Ovarian carcinoma in 13 • Breast cancer in 2 • Cervical cancer in 3 • Endometrial cancer in 2 • Carcinoma of the stomach in 4 • Carcinoma of the pancreas in 1 • Carcinoma of gall bladder in 1 • Carcinoma of the colon in 3 • Carcinoma of the kidney in 1 • Leukemia in 3 • Adenocarcinoma of unknown origin in 2 • Carcinoma of the rectum in 1 • Carcinoma of the bladder in 1 • Hodgkin's lymphoma in 1 • Haepatocellular carcinoma in 6	• Hepatocellular Carcinoma and cirrhosis of the liver in 1 • Carcinoma of the breast with liver metastases in 1 • Carcinoma of the stomach with liver metastases in 1

[Table/Fig-1]: The division of patients into the groups and subgroups with causes.

Variables	Group A subgroup 1 (n=28)	Group A subgroup 2 (n=18)	Group B subgroup 3 (n=44)	Group B subgroup 4 (n=3)	Group A (subgroup 1+2) (n=46)	Group A (subgroup 3+4) (n=47)
1. Cholesterol (mg/dL)						
Mean±SD	13.29± 5.73	26.68± 18.81	99.11± 11.58	71.67± 5.51	18.37± 13.95	97.36± 13.14
Median	12.5	20.5	99.0	69.0	13.0	97.0
Range	4.0-26.0	6.0-69.0	69.0-123.0	68.0-78.0	4.0-69.0	68.0-123.0
2. Fibronectin (ng/mL)						
Mean±SD	8.78± 3.99	13.39± 10.72	67.27± 20.01	30.48± 5.09	10.59± 7.63	64.93± 21.41
Median	7.905	8.9	67.905	30.08	8.115	66.78
Range	1.61-14.96	1.61-36.2	36.3-100.0	25.6-35.76	1.61-36.2	25.6-100.0
3. Total protein (g/dL)						
Mean±SD	1.81± 0.56	1.68± 0.29	1.96± 0.63	1.45± 0.47	1.76± 0.47	1.93± 0.63
Median	1.80	1.74	1.80	1.20	1.76	1.80
Range	1.07-3.33	1.16-2.32	1.14-3.34	1.16-1.99	1.07-3.33	1.14-3.34

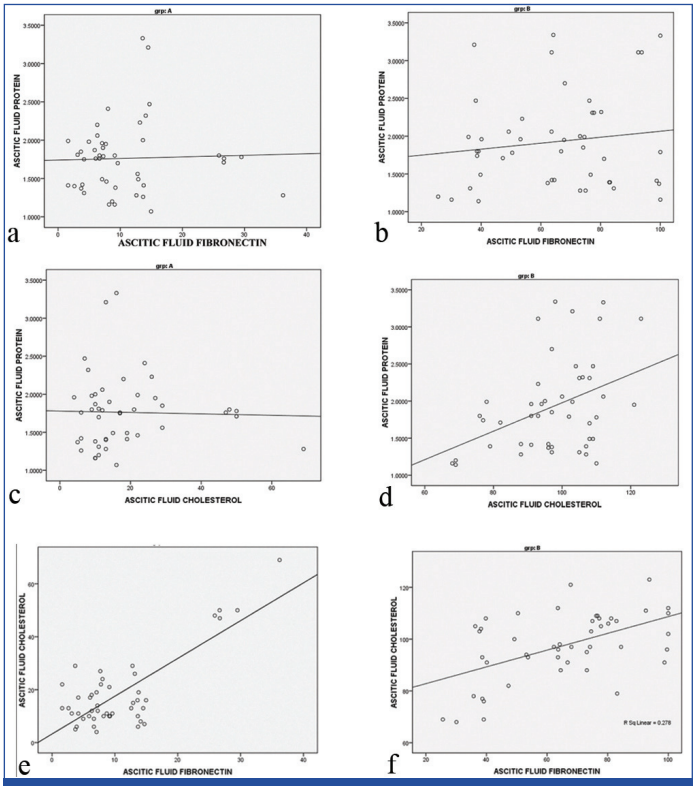
[Table/Fig-2]: Ascitic fluid concentrations of cholesterol, fibronectin, and protein.

64.93±21.41 ng/mL, while it was 10.59±7.63 ng/mL in non malignant ascites (highly significant with p<0.001). The mean ascitic fluid cholesterol level for patients with malignant ascites was 97.36±13.14 mg/dL, while it was 18.37±13.95 mg/dL for patients with non malignant ascites (highly significant with p<0.001). The mean SACG was 90.19±31.30 mg/dL for malignant ascites and 124.93±50.55 mg/dL for non malignant ascites (highly significant with p<0.001) [Table/Fig-3]. In the present study, the difference in the proportion of Ascitic Fluid Total Protein (AFTP) values between non malignant ascites and malignant ascites was statistically insignificant (p-value >0.05).

Parameters	Group A (mean±SD)	Group B (mean±SD)	p-value
Ascitic fluid cholesterol (mg/dL)	18.37±13.95	97.36±13.14	0.0001
Ascitic fibronectin (ng/mL)	10.59±7.63	64.93±21.41	0.0001
Ascitic fluid protein (mg/dL)	1.76±0.47	1.93±0.63	0.290
SAAG (g/dL)	2.40±0.68	2.48±0.71	0.509
SACG (mg/dL)	124.93±50.55	90.19±31.30	0.001

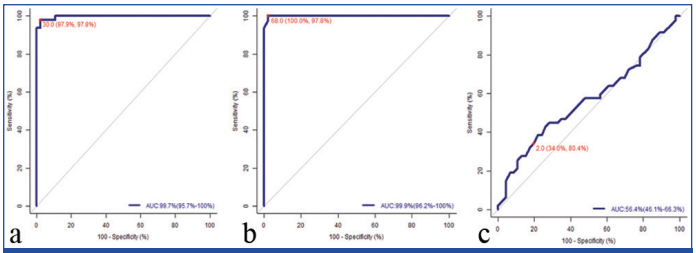
[Table/Fig-3]: The analytes in ascitic fluid in both malignant and non malignant ascites and their p-values.
*p-value <0.05 is significant. Mann-Whitney U test; SAAG: Serum ascites albumin gradient; SACG: Serum ascites cholesterol gradient

There was a positive significant correlation between ascitic fibronectin and cholesterol in both non malignant and malignant ascites groups and a positive insignificant correlation between ascitic fibronectin and protein in both non malignant and malignant ascites groups. There is a positive significant correlation between ascitic cholesterol and protein in the malignant ascites group. The correlation of ascitic fluid concentrations of cholesterol and fibronectin tended to be slightly better than that of any parameter with ascitic protein concentration, and this difference was statistically significant [Table/Fig-4a-f].

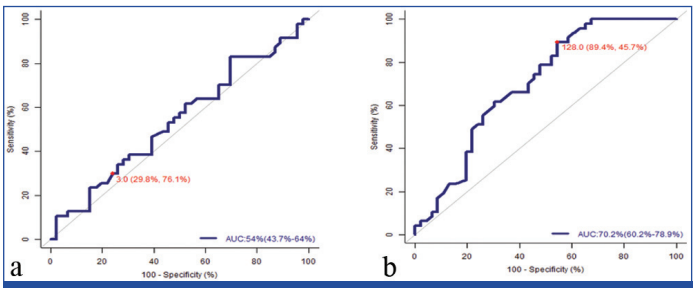


[Table/Fig-4]: Linear regression showing correlation between ascitic fluid cholesterol, fibronectin and total protein. a) Positive insignificant correlation between ascitic fibronectin and protein in non malignant ascites group. Correlation coefficient (r) is 0.011, p-value=0.941 (*p-value <0.05 is significant). b) Positive insignificant correlation between ascitic fibronectin and protein in malignant ascites group. Correlation coefficient (r) is 0.062, p-value=0.677 (*p-value <0.05 is significant). c) Negative insignificant correlation between ascitic cholesterol and protein in non malignant ascites group. Correlation coefficient (r) is -0.081, p-value=0.592 (*p-value <0.05 is significant). d) Positive significant correlation between ascitic cholesterol and protein in malignant ascites group. Correlation coefficient (r) is 0.365, p-value=0.012* (*p-value <0.05 is significant). e) Positive significant correlation between ascitic fibronectin and cholesterol in non malignant ascites group. Correlation coefficient (r) is 0.303, p-value=0.041* (*p-value <0.05 is significant). f) Positive significant correlation between ascitic fibronectin and cholesterol in malignant ascites group. Correlation coefficient (r) is 0.478, p-value=0.001* (*p-value <0.05 is significant).

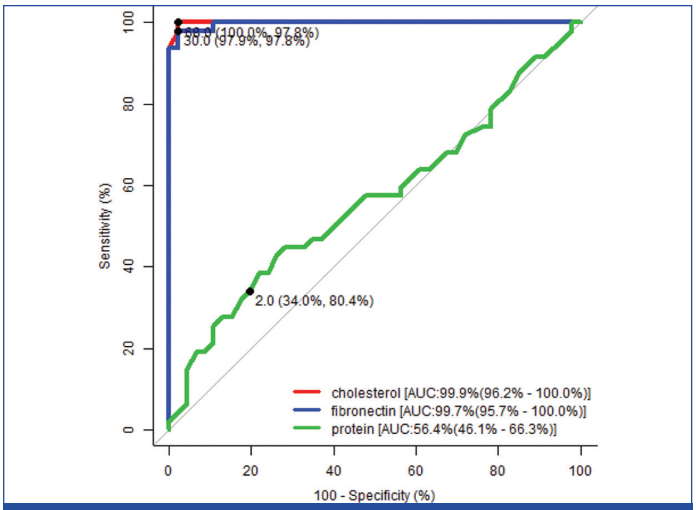
As demonstrated by the ROC [Table/Fig-5-7], the differential diagnostic effectiveness of cholesterol and fibronectin was better than that of protein in distinguishing between non malignant ascitic patients and malignant ascitic patients.



[Table/Fig-5]: ROC in ascitic fluid. Ascitic fibronectin (a) Ascitic cholesterol (b) Ascitic total protein (c).



[Table/Fig-6]: ROC for SAAG (a); and SACG (b).



[Table/Fig-7]: Receiver Operating Characteristics (ROC) showing sensitivity and specificity at different discrimination levels for cholesterol, fibronectin and protein. As differential diagnostic efficiency progress, the curve approaches the left upper corner (100% sensitivity and 100% specificity) of the figure.

The present analysis was conducted by measuring sensitivity, specificity, positive and NPV, and diagnostic efficiency [Table/Fig-8]. These values were obtained after plotting the ROC curves.

Parameters	Ascitic fluid cholesterol	Ascitic fluid fibronectin	Ascitic fluid total protein	SAAG	SACG
Sensitivity (%)	100	97.87	34.04	29.78	89.36
Specificity (%)	97.82	97.82	80.43	76.08	45.65
Youden index	0.978	0.957	0.145	0.059	0.35
NPV (%)	100	97.83	54.41	51.47	80.77
PPV (%)	97.92	97.87	64.00	56.00	62.69
Accuracy (%)	98.92	97.85	56.99	52.69	67.74
AUC	0.999 (0.962, 1)	0.997 (0.957, 1)	0.564 (0.461, 0.663)	0.54 (0.437, 0.64)	0.70 (0.602, 0.79)
p-value	0.001*	0.001*	0.146	0.255	0.001*
Cut-off value	68 mg/dL	30 ng/mL	2 g/dL	3 g/dL	128 mg%

[Table/Fig-8]: The sensitivity, specificity, Youden index, Negative Predictive Value (NPV), Positive Predictive Value (PPV), accuracy, area under curve and p- values of the ascitic fluid analytes at various cut-off values.
*p-value <0.05 is significant. SAAG: Serum ascites albumin gradient; SACG: Serum ascites cholesterol gradient; AUC: Area under the curve

Using various cut-off values, the diagnostic accuracy of ascitic fluid cholesterol, fibronectin, total protein, SAAG, and SACG in differentiating malignancy-related ascitic fluid from non malignant ascites were determined to be 98.92%, 97.85%, 56.99%, 52.69%, and 67.74%, respectively [Table/Fig-8].

DISCUSSION

The mean age of the study participants was 52.25±12.74 years, with the majority of participants being over 50 years old. This finding is similar to a study conducted by Kumar B et al., where the mean age of participants was 51.5 years [21]. Other studies by Joshi R et al., and Khan FY et al., also reported mean ages of ascites patients that were similar to this study, while studies by Muhie OA and Mehra D et al., reported lower mean ages [22-25]. In the present study, ascitic fluid cytology was positive in 70.2% of patients with malignancies. This is much higher compared to the results of Malabu UH et al., where cytology was positive in 22.7% of malignancy-related ascites cases [26]. This difference may be attributed to variations in the number of cases of peritoneal carcinomatosis in the two studies. Castaldo G et al., reported a sensitivity of 40%-60% for cytology in their article [27]. The sensitivity of ascitic fluid cytology is generally lower in overall malignancy-related ascites compared to peritoneal carcinomatosis, as not all cases are associated with peritoneal carcinomatosis [28].

In the present study, the difference in the proportion of AFTP values between non malignant and malignant ascites was statistically insignificant (p-value >0.05), with an accuracy of 57%. This is lower than the findings of Ekpe EEL et al., Ekpe L, Ekpe EEL and Ebughe GA, Deverbizier G et al., Sastry AS et al., and Sood A et al., where the diagnostic accuracy of AFTP ranged from 62.5% to 85% [8,13,17,29-31]. However, it is higher than the finding of Ekpe L, who reported a diagnostic accuracy of AFTP of 39% [18]. In the present study, SAAG value was 2.40±0.68 g/dL in non malignant ascites and 2.48±0.71 g/dL in malignant ascites (p>0.05), with an accuracy of 67.74%. These findings are similar to the results of Ekpe EEL and Ebughe GA, Lee CM et al., Sharatchandra LK et al., Runyon BA et al., Laudano OM et al., and Nadeem MA et al., [17,32-36].

In the present study, the ascitic fluid cholesterol and mean SACG were significantly higher in malignant ascites compared to non malignant ascites (p<0.001, statistically highly significant). Prieto M et al., found that ascitic fluid cholesterol concentrations were significantly elevated in patients with peritoneal metastases and were more effective than AFTP, LDH, and SAAG in differentiating ascites due to liver disease [37]. Similarly, Sharatchandra LK et al., reported similar findings, with SACG values in cirrhosis, tuberculosis, and malignancies of 99.2±27.8, 54.16±36.26, and 50±23 mg/dL, respectively, and a sensitivity of 80% [34]. Another study by Bjelakovi G et al., also found that cholesterol levels were significantly higher in malignant ascites compared to cirrhotic ascites [38]. This may be due to increased movement of plasma lipoproteins into the peritoneal cavity [39]. Additionally, the current study found that at a cut-off level of 68 mg/dL, cholesterol in ascitic fluid has a sensitivity of 100%, specificity of 97.8%, PPV of 97.9%, NPV of 100%, and diagnostic accuracy of 98.9% in distinguishing malignant from non malignant ascites. At a cut-off level of 128 mg/dL, SACG has a sensitivity of 89.36%, specificity of 45.6%, PPV of 62.7%, NPV of 80.8%, and diagnostic accuracy of 67.74% in differentiating malignant from non malignant ascites. These findings are similar to the study by Vyakaranam S et al., which reported that ascitic fluid cholesterol with a critical value of >62 mg/dL had a diagnostic accuracy of 96% [18]. In the study by Vyakaranam S et al., SACG with a cut-off value of 53 mg% achieved a diagnostic accuracy of 94% in distinguishing malignant ascites from cirrhotic and tuberculous ascites, which is higher than the results of the present study [40]. Rana SV et al., found that ascitic fluid cholesterol with a cut-off value of 70 mg/dL had a specificity of 100% and sensitivity of 96% in identifying malignancy, which is consistent with the findings of the present study [40]. Other studies by Sharatchandra LK et al.,

Ranjith D et al., Rana SV et al., Gupta R et al., and Laudano OM et al., also support the diagnostic accuracy of ascitic fluid cholesterol in malignancy identification [33,35,40-42]. However, the findings of this study regarding SACG are contrary to the results of Sastry AS et al., and Sapra V et al., whose studies reported sensitivities and accuracies of 94% and 90%, respectively [30,43]. The results of the present study align with the study by Dharwadkar K et al., which reported an accuracy of 68% and sensitivity of 60.4% at a cut-off value of less than 95 mg/dL for SACG [19].

Fibronectin was able to differentiate malignant from non malignant ascites ($p<0.001$) with a diagnostic accuracy of 97.8%. This is higher than the findings of Aksoy H et al., Khan FY et al., Gerbes AL et al., Ekpe L et al., and Ekpe EEL et al., where diagnostic accuracy ranged from 80% to 94.7% [6,8,9,13,44]. These results are consistent with the studies by Lee CM et al., Ghilain JM et al., and Scholmerich J et al., which also reported high accuracy for fibronectin in ascitic fluid [32,45,46].

Sood A et al., found that elevated concentrations of ascitic fibronectin were significantly higher in malignancy-associated ascitic fluid compared to non malignant ascitic fluid [31]. They established a correlation between malignancy and fibronectin levels. In the present study, diagnostic accuracy of fibronectin in ascitic fluid was determined to be 97.8%, using a cut-off value of 30 ng/mL. The sensitivity, specificity, and accuracy of fibronectin were 97.9%, 97.8%, and 97.8%, respectively. These findings are comparable to the study by Sood A et al., who reported an accuracy of 97.1% and a sensitivity of 100% [31]. Ghilain JM et al., reported a slightly lower diagnostic accuracy of 85% [46]. Lee CM et al., also found a diagnostic accuracy of 95.9% for ascitic fibronectin [32]. Siddiqui RA et al., reported 100% accuracy for fibronectin compared to 78.7% for malignant cytology [11]. TThe specificity of ascitic fibronectin in the present study (98%) reported by Prieto M et al., but lower than the 100% accuracy reported by Scholmerich J et al., and (97.8%) is similar to that reported by Colli A et al., (93%) and higher than the 88% reported by Gerbes AL and Archimandritis et al., reported lower than the 100% accuracy [37,46-49]. It should be noted that the cut-off levels differed among the five studies mentioned.

Limitation(s)

The present study may be limited by the fact that the sample size was rather small. Using a larger sample size would be needed to validate these findings. Additionally, the study was conducted at a single centre. A multicentre study would have provided more robust results.

CONCLUSION(S)

Based on the data and findings in the present study, it is evident that ascitic fluid levels of cholesterol and fibronectin are effective in differentiating between malignant and non malignant ascites. Fibronectin exhibits greater sensitivity in diagnosing malignancy compared to cytology, allowing for early initiation of therapy before all final results are obtained for further management. Ascitic fluid fibronectin may serve as a tumour marker in distinguishing between malignant and non malignant ascites. Since cholesterol can be easily and economically estimated, it could be recommended as a first-line parameter for ascitic fluid analysis. However, further prospective studies with a larger number of subjects are necessary to validate these findings.

Author's contribution: Dr. Deebyendu Sahu was the primary investigator and responsible for data collection. Dr. Sarata Chandra Singh contributed to the study design and manuscript writing. Dr. Bibhu Prasad Behera, Dr. Thakura Soren, and Dr. Tanmaini Das were involved in manuscript writing, statistical analysis, data interpretation. Dr. Bibhu Prasad Behera is the corresponding author.

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